2.2.14 Numerical Modeling and Fluids Experiments for Materials Processing and Protein Crystallization

The presentation highlights two of the three NASA funded projects currently in progress under this task:

1. Principal Investigator: Study of Fluid Flow Control in Protein Crystallization Using Strong Magnetic Fields. CoIs: F. Leslie and E. Ciszak - Fluid physics NRA

2. Co Investigator: Fundamental Studies of Crystal Growth of Microporous Materials, Flight definition research grant with Dr. Prabir Dutta, PI, The Ohio State University

Included here is a brief outline of the protein crystallization project (Co Investigator: Crystal Growth of ZnSe and related Ternary Compound Semiconductors by Vapor Transport with Dr. Ching-Hua Su, PI, NASA MSFC).

Introduction: An important component in biotechnology, particularly in the area of protein engineering and rational drug design is the knowledge of the precise three-dimensional molecular structure of proteins. The quality of structural information obtained from X-ray diffraction methods is directly dependent on the degree of perfection of the protein crystals. As a consequence, the growth of high quality macromolecular crystals for diffraction analyses has been the central focus for biochemists, biologists, and bioengineers.

Macromolecular crystals are obtained from solutions that contain the crystallizing species in equilibrium with higher aggregates, ions, precipitants, other possible phases of the protein, foreign particles, the walls of the container, and a likely host of other impurities. By changing transport modes in general, i.e., reduction of convection and sedimentation, as is achieved in "microgravity", we have been able to dramatically affect the movement and distribution of macromolecules in the fluid, and thus their transport, formation of crystal nuclei, and adsorption to the crystal surface. While a limited number of high quality crystals from space flights have been obtained, as the recent National Research Council (NRC) review of the NASA microgravity crystallization program pointed out, the scientific approach and research in crystallization of proteins has been mainly empirical yielding inconclusive results [1].

Hypothesis: We postulate that we can reduce convection in ground-based experiments and we can understand the different aspects of convection control through the use of strong magnetic fields and field gradients. We postulate that limited convection in a magnetic field will provide the environment for the growth of high quality crystals. The approach exploits the variation of fluid magnetic susceptibility with concentration for this purpose and the convective damping is realized by appropriately positioning the crystal growth cell so that the magnetic susceptibility force counteracts terrestrial gravity.

Objective: The general objective is to test the hypothesis of convective control using a strong magnetic field and magnetic field gradient and to understand the nature of the various forces that come into play. Specifically we aim to delineate causative factors and to quantify them through experiments, analysis and numerical modeling. Once the basic understanding is obtained, the study will focus on testing the hypothesis on proteins of pyruvate dehydrogenase complex (PDC), proteins E1 and E3.

Obtaining high crystal quality of these proteins is of great importance to structural biologists since their structures need to be determined. Our approach will simultaneously provide the first implementation of our flow control hypotheses and investigation.

Specific goals for the investigation are:

1. To develop an understanding of convection control in diamagnetic fluids with concentration gradients through experimentation and numerical modeling. Specifically solutal buoyancy driven convection due to crystal growth will be considered.
2. To develop predictive measures for successful crystallization in a magnetic field using analyses and numerical modeling for use in future protein crystal growth experiments. This will establish criteria that can be used to estimate the efficacy of magnetic field flow damping on crystallization of candidate proteins.
3. To demonstrate the understanding of convection damping by high magnetic fields to a class of proteins that is of interest and whose structure is as yet not determined.
4. To compare quantitatively, the quality of the grown crystals with and without a magnetic field. X-ray diffraction techniques will be used for the comparative studies.

Value: The proposed work is ground-based with both theoretical and experimental investigation of crystallization phenomena with emphasis on the important fluid physics element of the protein crystallization process. Further, the proposed investigation employs an interdisciplinary approach to advance the research in the important area of biocrystallization and thus biotechnology. Understanding the fluid flow due to solutal buoyancy convection and controlling that flow through the application of a strong magnetic field and field gradient are important objectives. This implies obtaining quantitative information through carefully conducted experiments, developing predictive capability through analytical and numerical simulations, and applying it to proteins whose structures are as yet undetermined. The studies will directly provide a predictive tool and a technique to further reduce convective contamination in space experiments on board the *ISS*.

Our research will determine whether the use of a strong magnetic field has significant impact on the crystallization of the most challenging macromolecular systems. Extending our success in crystallization in a magnetic field with proteins from arguably the most exciting and challenging multienzyme assembly of the human body, i.e. pyruvate dehydrogenase complex will certainly provide a breakthrough for macromolecular crystal growers.

[1.] National Research Council, Space Studies Board, Commission on Physical Sciences, Mathematics and Applications, in Future Biotechnology Research on the International Space Station, national Academy Press, Washington, DC., 2000, 10-16.

For further information, see the presentation made by Dr. Narayanan Ramachandran in Appendix B, page M-184.